4568
Topic Category: 4071-ASIP Liver fibrosis and cirrhosis

First Author: Komal Ramani
Cedars-Sinai Medical Center 8700 Beverly Blvd Los Angeles, CA 90048
United States
Phone:
komal.ramani@cshs.org

First Author is a: Investigator
First Author is a member of: American Society for Investigative Pathology
First Author Degree: PhD, DSc, or equivalent

Presentation Preference: Oral

Sponsor: Komal Ramani
Sponsor Phone: 5629658303
komal.ramani@cshs.org
Sponsor's Society: Pathology - American Society for Investigative Pathology (ASIP) - Host Society
Keywords: 1. Alcoholic liver disease 2. Hepatic stellate cells 3. Liver fibrosis

A-Kinase Anchoring Protein Phosphorylation as a Therapeutic Target for Alcohol Liver Injury

Komal Ramani[1], Maria Lauda Tomasi[1], Joshua Berlind[1], Nirmala Mavila[1], Zhaolo Sun[2]. [1]Cedars-Sinai Medical Center, Los Angeles, CA, [2]Johns Hopkins University School of Medicine, Baltimore, MD

Hepatic stellate cells (HSC) activation associated with up-regulation of type I collagen is a distinct molecular response in alcoholic liver diease (ALD). A-kinase anchoring protein 12 (AKAP12) is a scaffold protein for protein kinases A/C (PKA/PKC) and cyclin-D1 that suppresses oncogenic proliferation, chemotaxis and cellular senescence. The scaffolding activity of AKAP12 is altered by site-specific phosphorylation that leads to loss of its interaction with PKC and cyclin-D1 allowing release of cyclin-D1 for cell cycle progression. AKAP12 tyrosine phosphorylation alters its binding to actin favoring cell motility. AKAP12 expression is induced during HSC activation but its scaffolding functions in liver function are undescribed so far. We identified increased levels of AKAP12 phosphorylation in ethanol-treated HSCs and hence hypothesized that this may be associated with altered scaffolding functions in alcohol-mediated HSC activation and liver injury. AKAP12 expression/interaction/phosphorylation was assayed in *in vitro* and *in vivo* ethanol models/human subjects by real-time PCR, co-immunoprecipitation-immunoblotting, phospho-proteomics/phosTag™/proximity ligation assays. Ethanol induced AKAP12 phosphorylation primarily in HSCs of mouse liver, but not hepatocytes. AKAP12 phosphorylation was also observed in alcoholic hepatitis/cirrhosis patient livers. AKAP12's scaffolding activity for PKC-α/cyclin-D1 was inhibited in ethanol-treated HSCs but not hepatocytes. AKAP12 acted as an anti-fibrogenic protein inhibiting HSC activation, however, ethanol reversed this anti-fibrogenic property through induction of AKAP12 phosphorylation. One mechanism of AKAP12's ability to control HSC activation was by its interaction with heat shock protein 47 (HSP47) that chaperones collagen and induces its secretion. Ethanol inhibited AKAP12-HSP47 and induced HSP47-collagen interaction. Ethanol-induced, phospho-AKAP12 adducts were unable to bind to HSP47 compared to its unphosphorylated counterpart, thereby proving that ethanol-mediated phosphorylation of AKAP12 inhibited the HSP47-AKAP12 scaffold. Silencing AKAP12 facilitated the chaperoning of collagen by HSP47. Hence, AKAP12 scaffolds HSP47 and regulates collagen-HSP47 interaction and this ability is quenched by ethanol through phosphorylation. The findings implicate important functions of AKAP12 in controlling HSP47/collagen maturation that is deregulated by its phosphorylation. By phospho-peptide mapping, we identified novel ethanol-responsive AKAP12 phospho-sites in human HSCs that may be further explored as therapeutic targets in ALD.

Support or Funding Information
Komal Ramani: (Applied Cell Biology and Physiology (ACB&P) division seed funding, Biomedical Sciences Department, Cedars-Sinai Medical Center, active) Komal Ramani (*5R00AA017774-05*, NIH, complete) Maria Lauda Tomasi: (*K01AA022372*, NIH, active) Nirmala Mavila (Applied Cell Biology and Physiology (ACB&P) division seed funding, Biomedical Sciences Department, Cedars-Sinai Medical Center) Zhaoli Sun (*R24AA025017*, NIAAA supported Clinical Resource for Alcoholic Hepatitis Investigations, Johns Hopkins University, active).